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Prediction of outcome in pre-dialysis renal failure. A 2 year prospective study utilising pregnancy associated plasma protein A as a marker

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Pregnancy associated plasma protein A (PAPPA) has recently been described in atherosclerotic plaques and elevated levels have been found in patients with acute coronary syndromes and coronary artery disease. The aim of this investigation was to evaluate the prognostic significance of PAPPA in a group of patients with chronic renal impairment who were not on renal replacement therapy.

Patients attending the pre-dialysis clinic (104 patients) were studied. A single serum sample was drawn at baseline and the patients followed prospectively for 2 years in order to determine outcome and cause of death. PAPPA was measured by an in-house ELISA method utilising commercially available antisera. For statistically analysis a PAPPA of > or equal to 0.01 mlU/ml was chosen. The results are shown below

<table>
<thead>
<tr>
<th>Cause of death</th>
<th>PAPPA &lt;0.01 mlU/ml (n=42)</th>
<th>PAPPA &gt;or = 0.01 mlU/ml (N=62)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Death</td>
<td>10 (24%)</td>
<td>11 (18%)</td>
</tr>
<tr>
<td>Cardiovascular events</td>
<td>9 (21%)</td>
<td>17 (27%)</td>
</tr>
<tr>
<td>Cause of death</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cardiac Arrest/MI/CCF</td>
<td>4 (10%)</td>
<td>4 (6%)</td>
</tr>
<tr>
<td>Stroke</td>
<td>2 (5%)</td>
<td>3 (5%)</td>
</tr>
<tr>
<td>Other</td>
<td>4 (10%)</td>
<td>4 (6%)</td>
</tr>
</tbody>
</table>

Further, Kaplan-Meier survival analysis failed to demonstrate significant differences in survival at the cut off chosen. In conclusion PAPPA was not useful as a predictor of outcome in patients with chronic renal impairment attending a pre-dialysis clinic

M24

Uregulation of Monocyte chemoattractant protein-1 in early stage diffuse scleroderma


Fibroblast derived monocyte chemoattractant protein-1 (MCP-1 or CCL2) is a candidate mediator that may link inflammatory and fibrotic processes in scleroderma (SSc) pathogenesis. This study examines the relationship between MCP-1 ligand and receptor expression and stage of scleroderma.

METHODS: Serum samples and skin biopsies were examined from 54 patients with SSc and 12 healthy controls. 20 had early (within 3 years from onset) diffuse cutaneous SSc (dcSSc), 14 late dcSSc and 20 limited cutaneous SSc (lcSSc). Levels of MCP-1 in serum were measured by commercial ELISA. Expression of MCP-1 and its major receptor CCR2 in snap-frozen skin biopsies was determined by immunohistochemistry. Fibroblasts were cultured from a randomly selected subgroup of early dcSSc and culture supernatants examined by western blot. Chemokine receptor expression (CCR2, CXCR2 and CCR5) on fibroblasts was determined by flow cytometry, comparing receptor-specific antibody binding with an isotype matched control.

RESULTS: MCP-1 serum levels (mean±sem) were significantly elevated in SSc patients (342±24 pg/ml) compared to controls (132±26 pg/ml, p=0.002) and were highest in the early dcSSc subset (433±47 pg/ml) compared to late dcSSc (307±41 pg/ml) and lcSSc (276±28 pg/ml). Western Blot analysis of cultured fibroblasts supernatants confirmed substantial overproduction (mean±sem) of MCP-1 by cells from early dcSSc (n=8) (RDU=37.5±1.2) compared with normal fibroblasts (RDU=8.3±2.0) (p=0.002). Immunohistochemistry showed strong expression of MCP-1 and CCR2 in skin from all patients with early stage dcSSc, but expression in late stage dcSSc and lcSSc was weaker or absent. By flow cytometry CCR2 was detectable only on early-stage dcSSc, both normal and SSc fibroblasts expressed CXCR2 whereas CCR5 was not detected.

CONCLUSION: High serum levels of MCP-1 in early diffuse disease, overproduction by fibroblasts cultured from early-stage dcSSc biopsies and CCR2 expression on fibroblasts confirm upregulation of the MCP-1 ligand-receptor axis in SSc and suggest that chemokine antagonism may be a logical therapeutic strategy.

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The role of graft expressed Fas-Ligand on allograft rejection

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Apoptosis acts as both a control and effector mechanism with in the immune system. The best-described receptor-ligand combination that induces this, comprises one member of the TNF receptor family, Fas, and one member of the TNF ligand family, Fas-ligand (FasL). This receptor/ligand combination contributes to immune privilege and acts as a homeostatic mechanism within the immune system. Pathological expression of FasL on certain tumour lines leads to immune evasion.

In the light of this evidence of modulation of the immune response by the expression of FasL, we hypothesised that graft parenchyma uses the expression of FasL as a protective mechanism in inflammation.

In a mouse model of skin transplantation, we have shown that FasL is upregulated during rejection, and that the presence of FasL leads to a prolongation of graft survival across a single (p=0.002) or multiple minor histocompatibility mismatches.

Considering the possible level at which FasL has its effect, there were many more infiltrating cells in the FasL+ than the wild type allograft. Using animals that had been primed with wild type skin, there was much more rapid rejection of FasL+ than wild type allografts. Suggesting that wild type grafts were afforded some degree of protection from the efferent limb of the immune response by the presence of FasL. This implies that FasL acts locally to reduce the immune response.

We looked at the afferent limb of the immune response in a cellular model. Wild type and FasL- male target cells were mixed with female control cells at a ratio of 1:1 and introduced intravenously into a female wild type recipient. The recipient mice then went on to receive a second inoculation of a 1:1 mixture of male and female wild type cell suspension. There was an increased rate of loss of the male cells when the recipients had been primed initially with FasL- cells, as well as an increase in the number of male antigen specific CD8+ cells identified with MHC peptide tetramer staining. This implies that the nature of the immune response induced varies to some degree on the basis of the FasL expression by the priming cell.

An understanding of the physiological role of FasL in inflammation will help us harness it as a modulatory agent in transplant biology.